

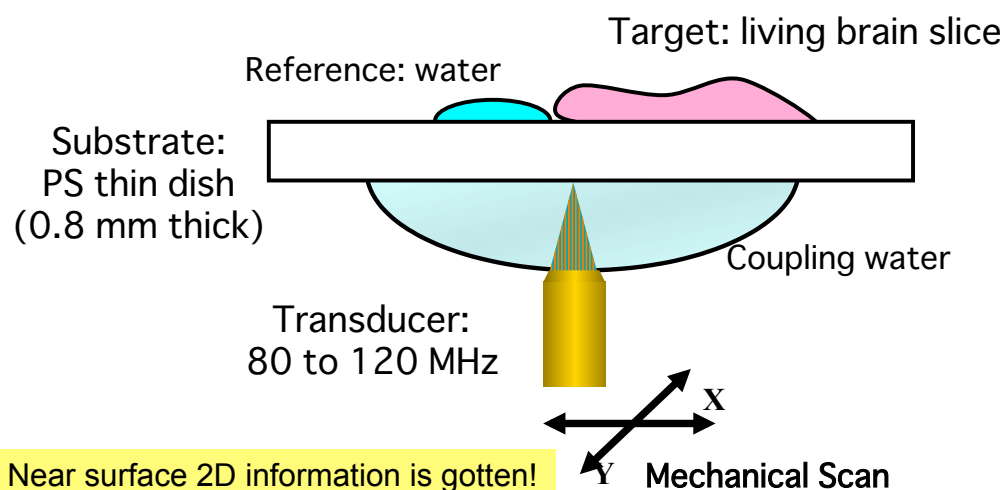
Where do the acoustic images come from? What are they? Where are they going?

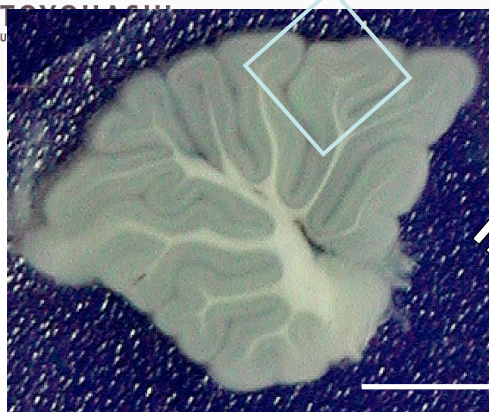
Sachiko Yoshida
Toyohashi University of Technology

Our targets: Living organs and cells of rat cerebellum

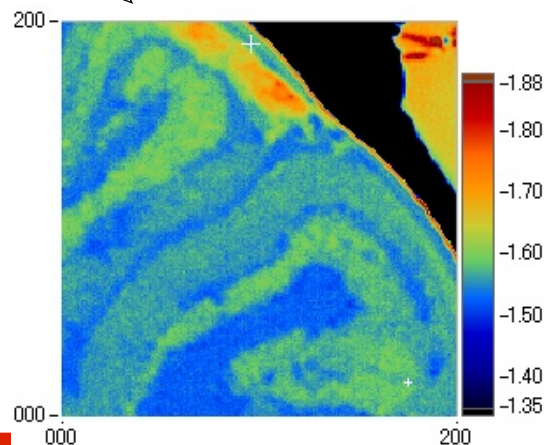
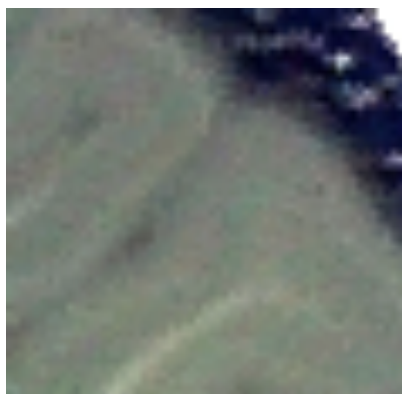
Materials: 400 μ m sliced rat cerebellar cortex and cultured glial cells

Methods: Acoustic impedance microscopy





Cerebellar slice observation
using middle frequency (80 – 120 MHz)



Optical image

Acoustic image

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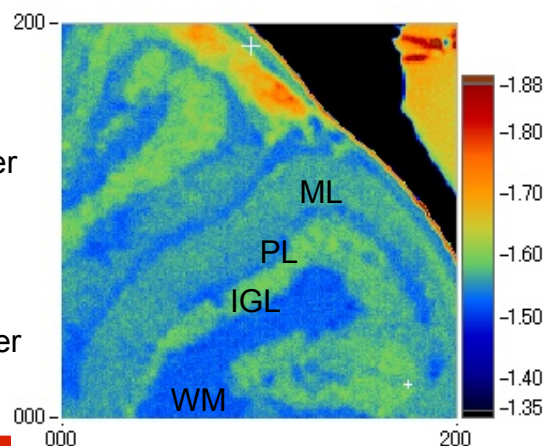
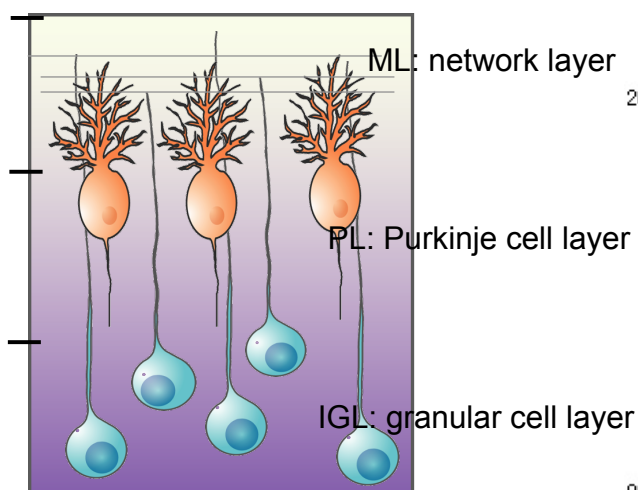


TOYOHASHI
UNIVERSITY OF TECHNOLOGY

Nuclei-rich layer: high (>1.58) impedance

Cytoskeleton-rich layer: middle (≈ 1.56) impedance

Lipid-rich layer: low (<1.54) impedance



Optical image

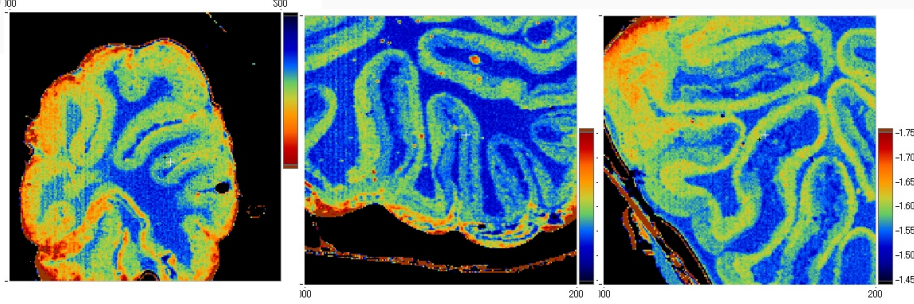
Acoustic image

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P7

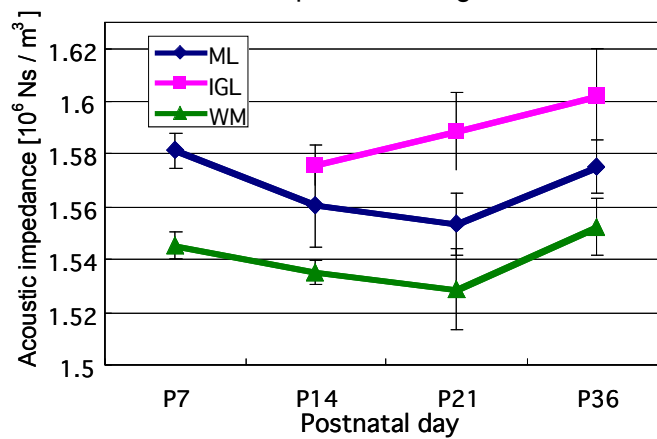
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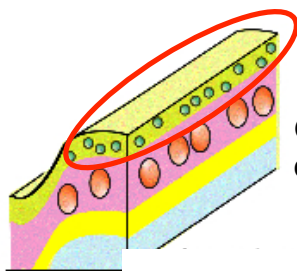


Acoustic impedance images

1mm

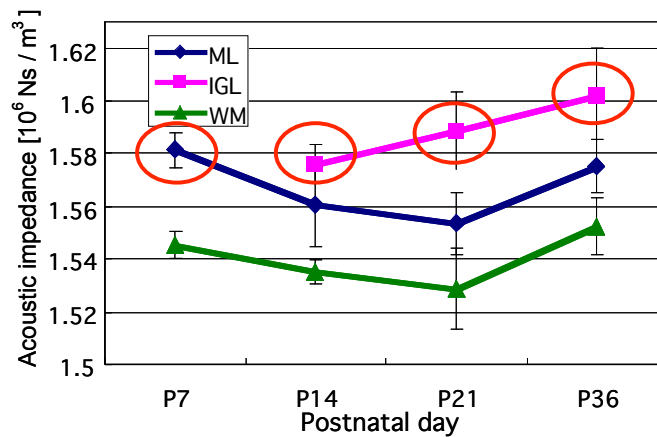
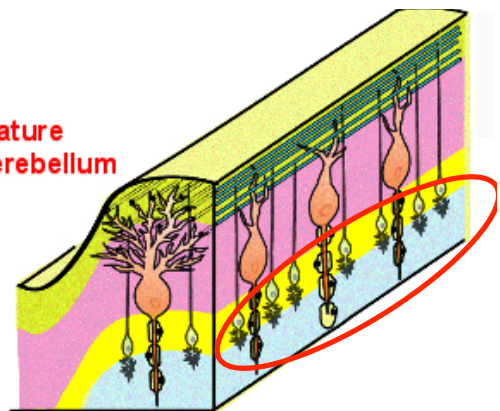


High impedance region moves to deep layer



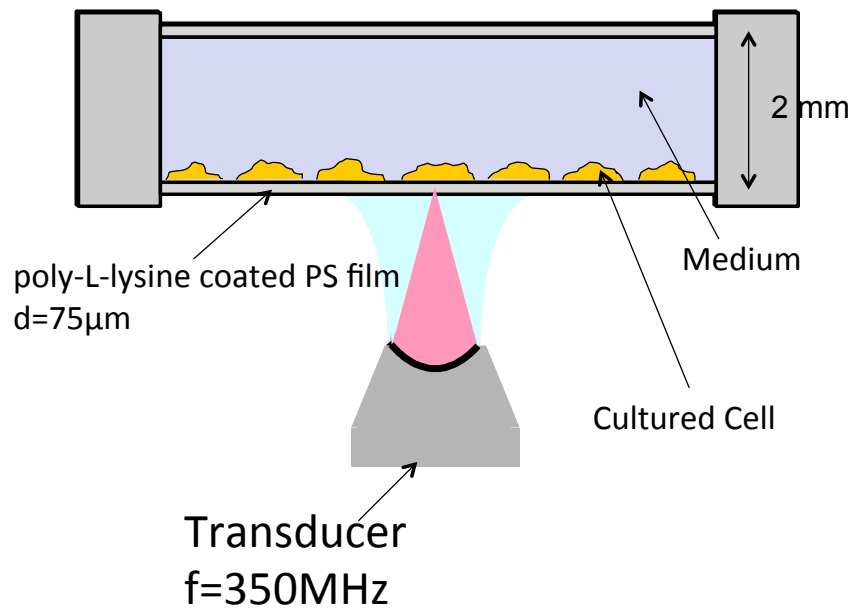
Granule cells move down to deep layer in development

Mature cerebellum

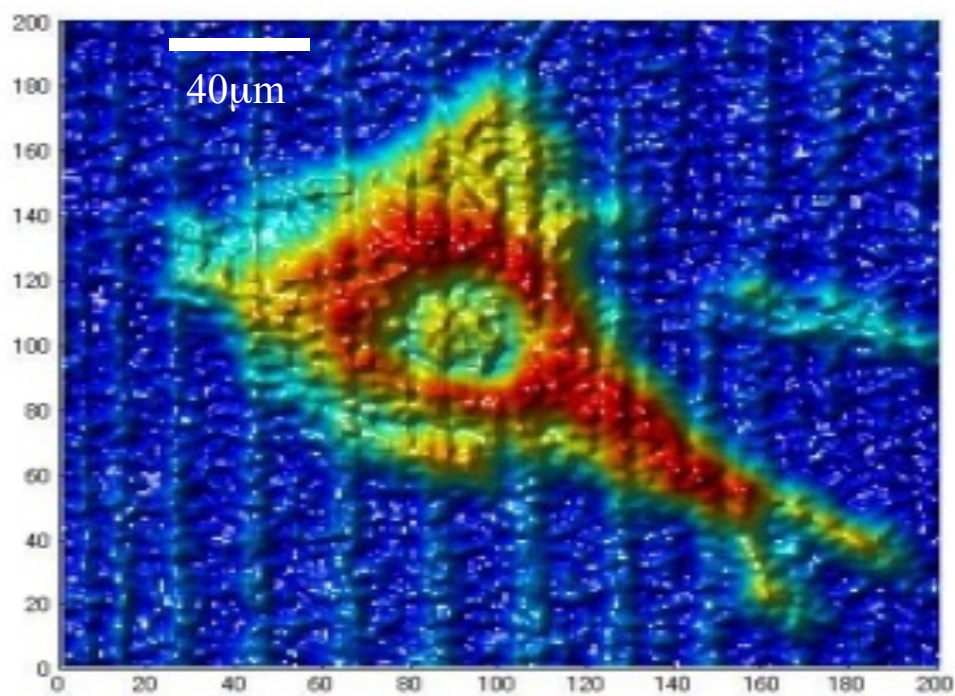


High impedance region moves to deep layer

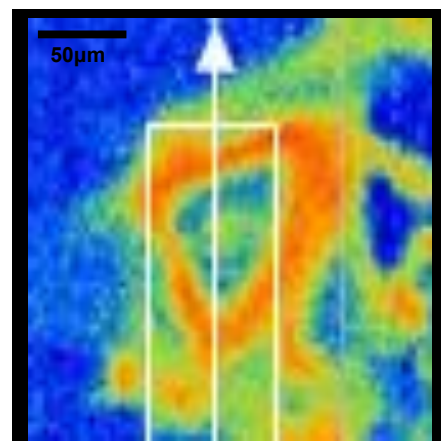
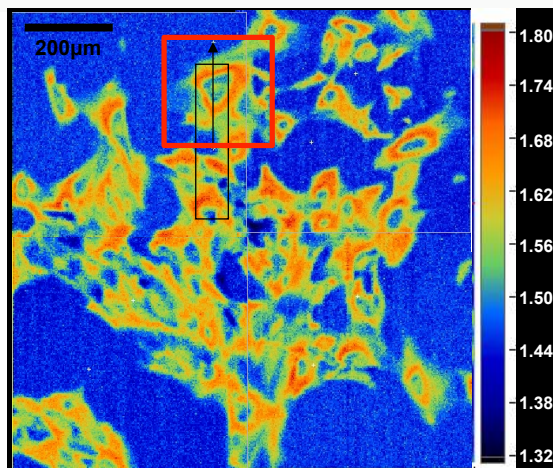
Subcellular observation



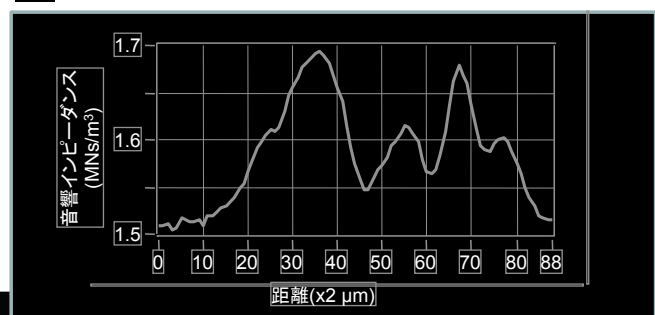
OptiCell



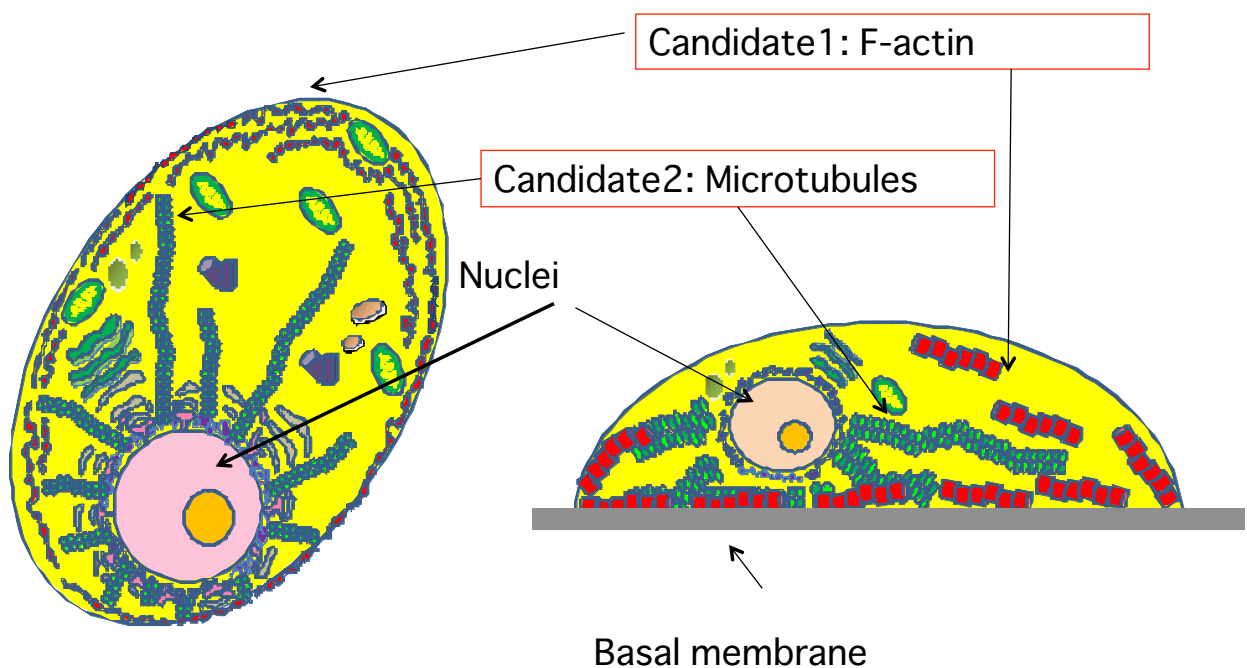
Acoustic image of a migrating glial cell

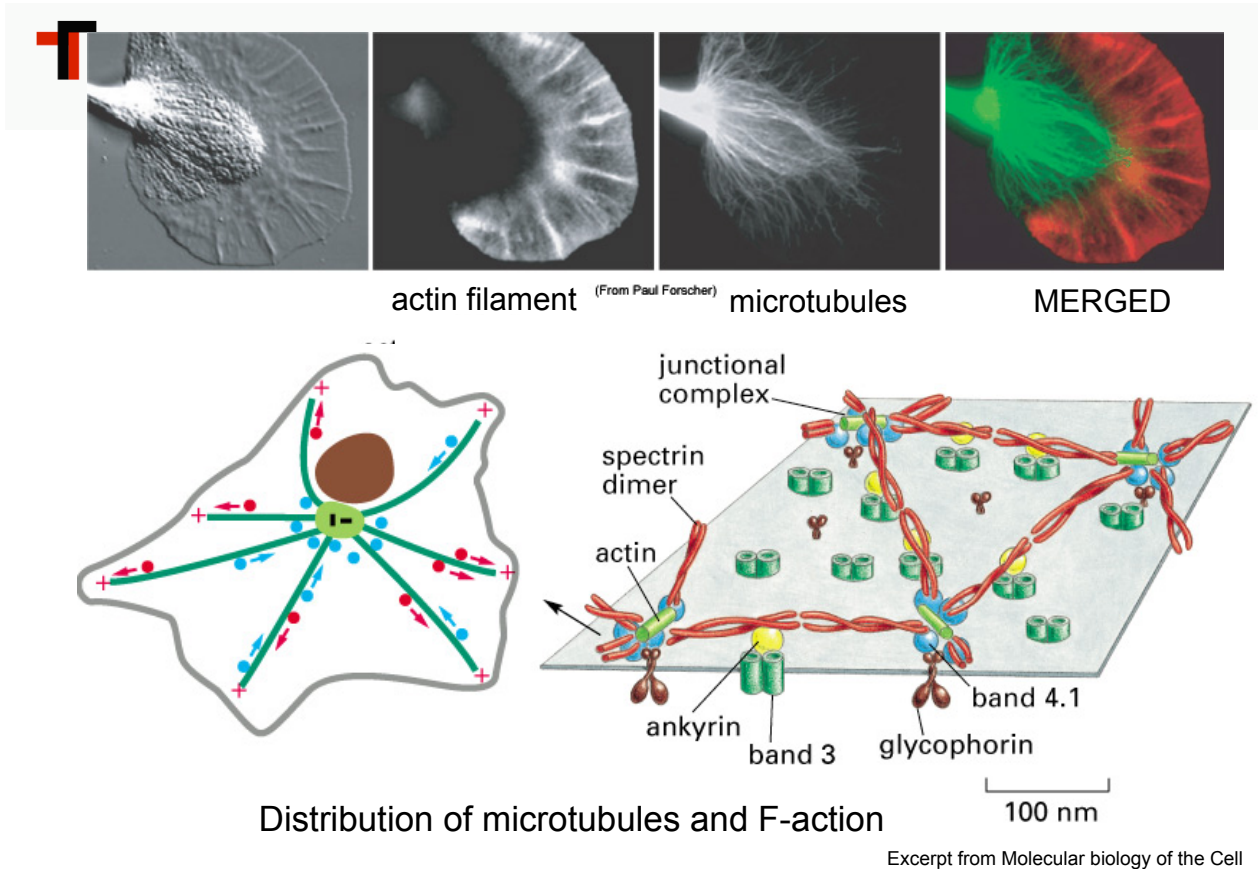


What is this organelle?

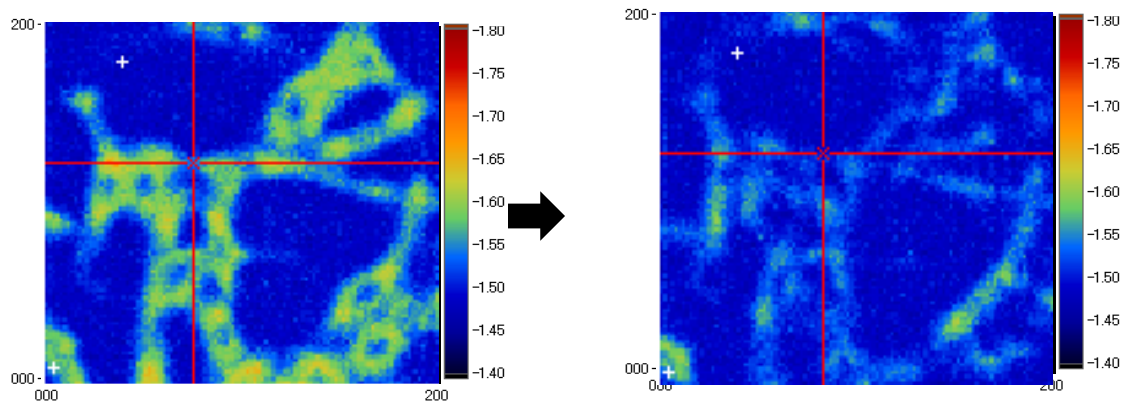
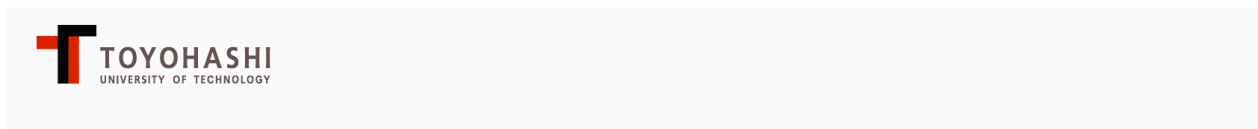


白矢に沿った音響インピーダンス



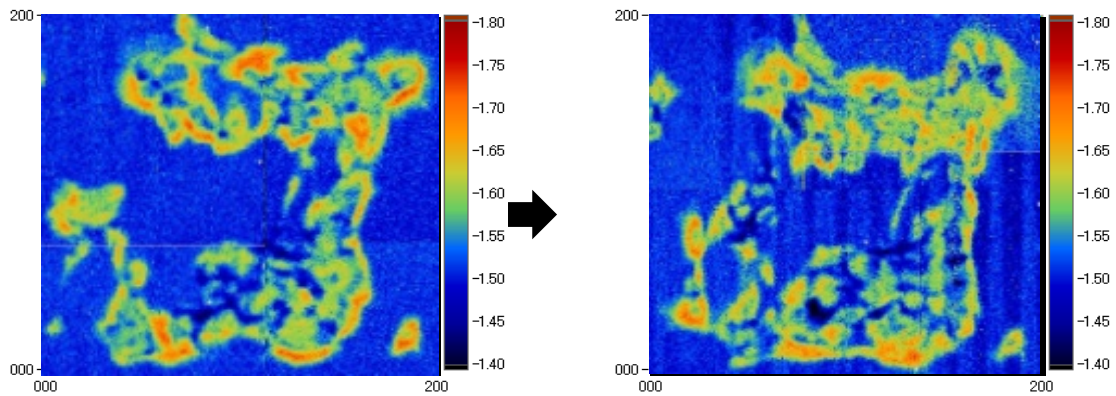


国立大学法人
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Depolymerization of F-actin decreased intracellular impedance

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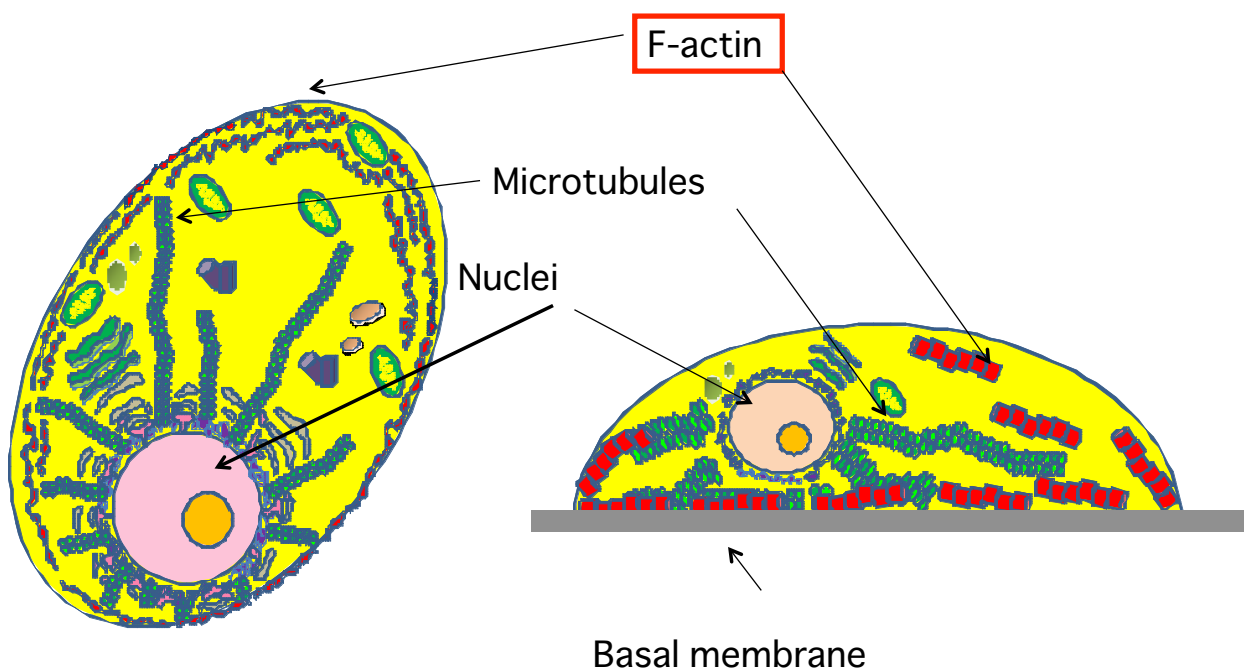


Living astrocyte

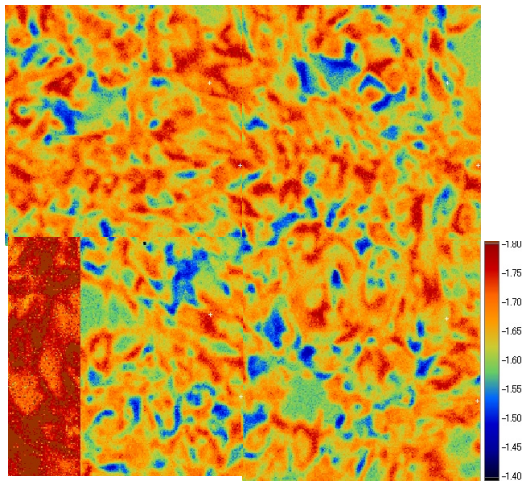
1 hr after Demecolcin application

Depolymerization of microtubules had little effect to intracellular impedance

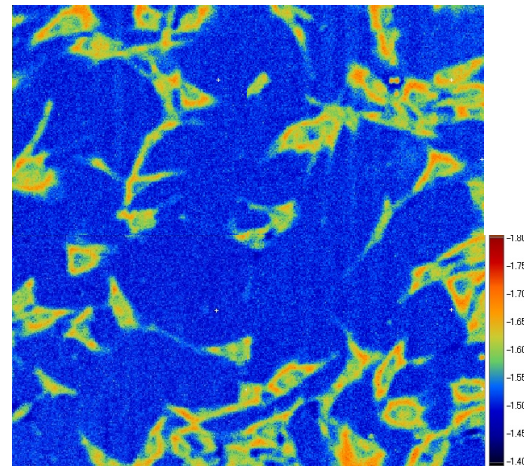
Result: Subcellular F-actin bundle is observed



Identification of a specific type of cells

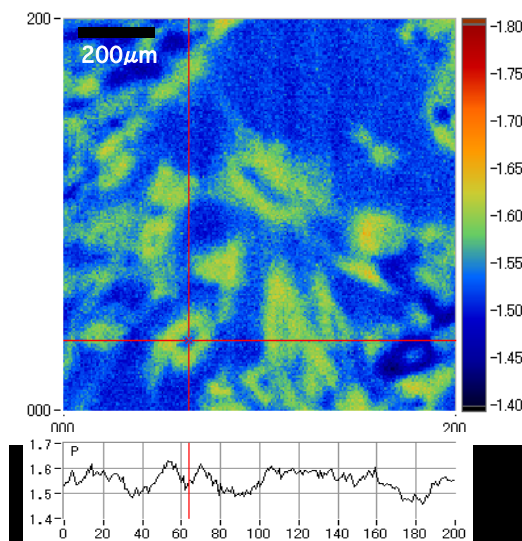


Wild type epidermal cells



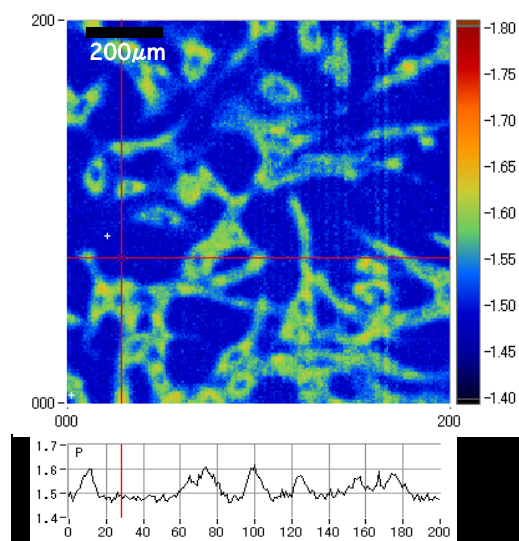
Immortalized epidermal cells

Astrocyte (normal cell)



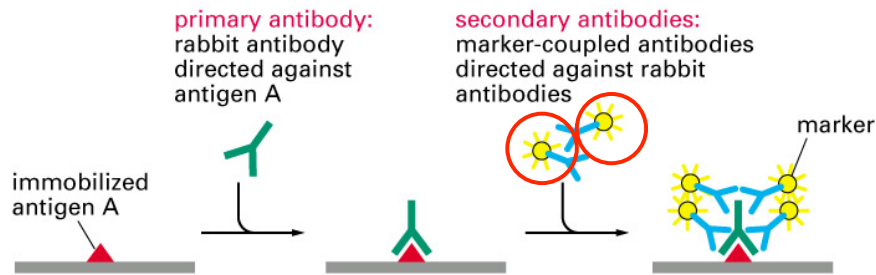
Ave. of Impedance 1.63

C6 Glioma (cancer cell)

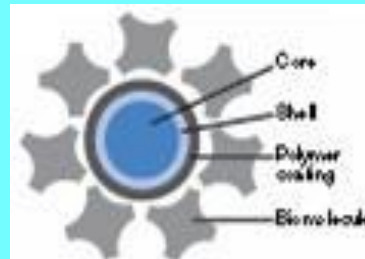


Ave. of Impedance 1.57

Experiment: **Acoustic immunohistochemistry** using Metal crystal

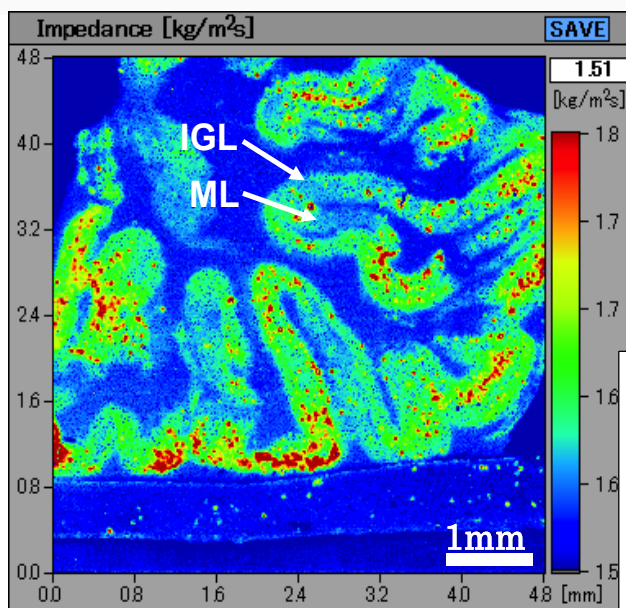


10~20nm diameter
core: semiconductor Cd
shell: Zinc sulfide
1 particle : 500 atoms

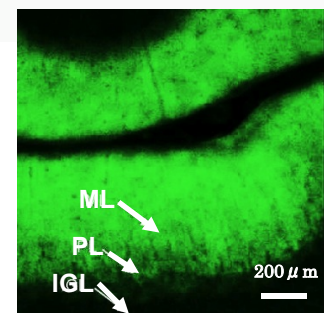


Q-Dot : Fluorescent heavy metal semiconductor nano particles

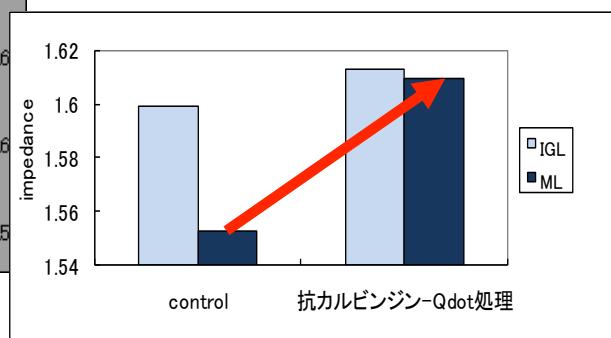
Primary antibody: Calbindin D28k



Acoustic staining image

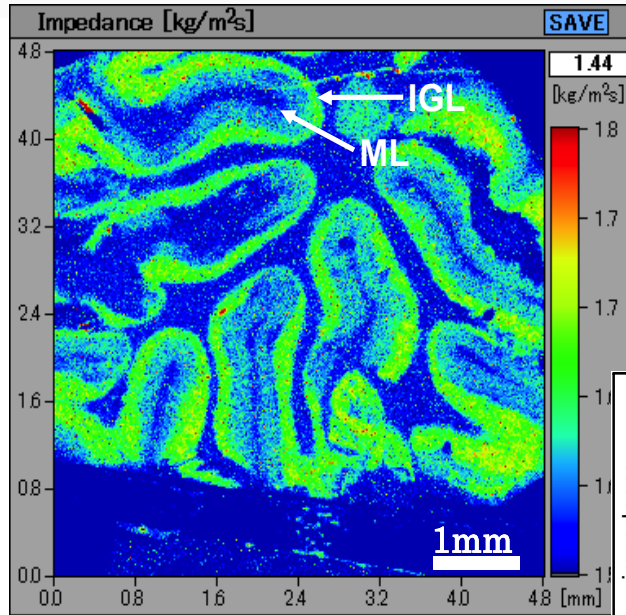


Fluorescent image

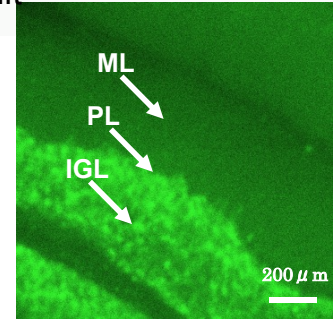


Impedance was increased by acoustic immunostaining to much proteins.

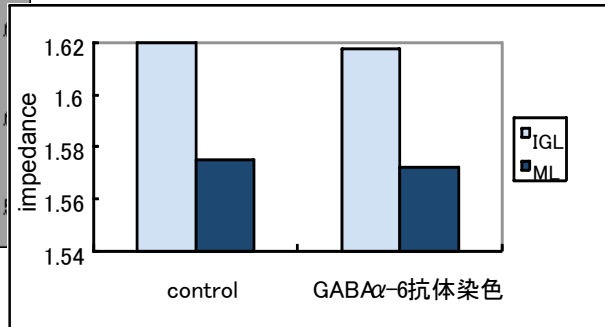
Primary antibody: GABAR α 6 subunit



Acoustic staining image



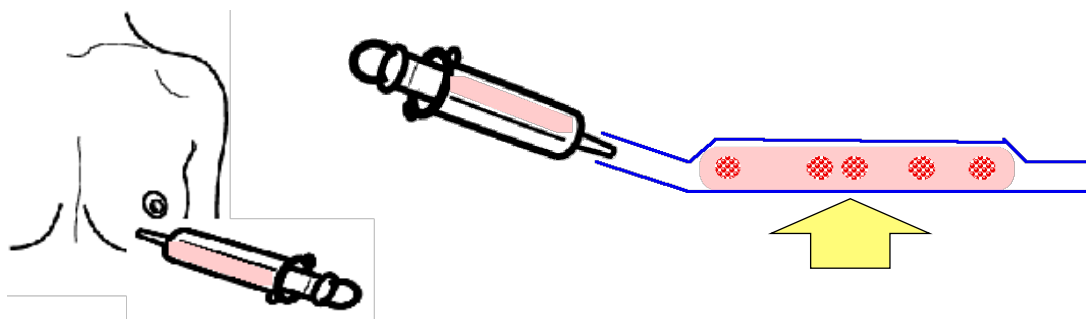
Fluorescent image



Impedance was not increased by acoustic immunostaining to less proteins.

In the future

Identification of biopsied tumor organ with attenuation observation



Biopsied sample is inserted to film cartridge and observed the density of nuclei, or distribution of specific markers (model)

Conclusions

- Middle level-reforming of substrate plate or film is effective to good acoustic impedance imaging.
- High freq. transducer makes cytoskeletal structure (F-actin) visualized.
- Surface protein in living cells are marked with Qdot or some heavy metal.

Thank you for your attention

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Kazuto Kobayashi

Hamamatsu University School of Medicine
Seiji Yamamoto

Toyohashi University of Technology
Many students

